# FLAVONOIDS OF ONAGRACEAE[1]

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ABSTRACT

Analyses of foliar flavonoids have been completed for approximately half of the species of Onagraceae and preliminary surveys have been carried out for at least one species of each of the 17 genera. Some 40 flavonoids, including glycoflavones, flavonols, and flavones, have been isolated and identified. Flavonols occur in all genera and in most species. Glycoflavones, either as the sole flavonoid constituent or co-occurring with flavonols, characterize the more generalized tribes, whereas only flavonols are found in the relatively specialized tribe Epilobieae. Glycoflavones, presumed primitive, occur in some members of all tribes except Fuchsieae and Epilobieae, whereas flavones occur only in Circaeeae, Fuchsieae, and Onagreae. In general, flavonoid data are strongly supportive of conclusions drawn from other lines of phylogenetic investigation about the relative degree of advancement of taxa, but they do not provide strong indications of phylogenetic relationship between genera or tribes.

Most studies of flavonoids in Onagraceae have concentrated on the compounds found in the leaves of these plants, and this paper will emphasize such studies. First, however, it seems worthwhile to review those studies that have been concerned with the floral pigments of this family. In the later sections of the paper, when dealing with foliar flavonoids, we shall not concern ourselves with anthocyanins. The anthocyanins of the flowers of *Fuchsia* and their influence on flower color have been investigated by several workers (Nozzolillo, 1969; Yazaki & Hayashi, 1967; Harborne, 1967; Crowden et al., 1977). The 3-glucosides and the 3,5-diglucosides of all six common anthocyanins are responsible for flower color in this genus. Variants in color appears to be almost wholly determined by these anthocyanins (Crowden et al., 1977). One of them, malvidin 3,5-diglucoside, also has been reported from the flowers of *Epilobium* and *Clarkia*, and another, cyanidin 3-glucoside, also from the flowers of *Clarkia* (Harborne, 1967). Dement and Raven (unpubl. data) examined the floral anthocyanins of 12 genera of the family. They found four widespread anthocyanins, and noted that several other anthocyanins seemed to have evolved independently in several lines. Cyanidin 3-glycoside was the most commonly encountered anthocyanin in Onagraceae, being present in *Calylophus, Gaura, Gongylocarpus, Hauya,* and *Xylonogra.*

A member of another class of floral pigments, the chalcone isosalipurposide, was reported as a floral pigment in 13 species from five genera of Onagraceae by Dement and Raven (1973, 1974). This chalcone is of particular interest because it represents a class of flavonoids that is apparently not found in the leaves of any Onagraceae. This chalcone plays an important ecological role in yellow-flowered Onagraceae, not reflecting light in the ultraviolet wavelengths so conspicuous in the portions of the same flowers where carotenoids predominate. Here it contributes to the formation of floral markings strikingly visible under ultraviolet light, and thus to the insects that visit the flowers of these plants.

At least one species from each genus and most subgenera within the family have been analyzed for its foliar flavonoids. Some 40 compounds have been isolated and identified from Onagraceae, not including anthocyanins. These include seven glycoflavones and six flavones. The remainder are flavonols based on kaempferol (5), quercetin (14), or myricetin (7). Most of the flavonols are 3-O-mono- and diglycosides with relatively few exhibiting seven or B-ring (3′ or 4′) substituents. At least four sulphated compounds, occurring in two different tribes, have been detected in the family. These data are summarized in Table 1. Base compounds and numbering systems are shown in Figure 1. Results for the individual tribes (summarized in Table 2) follow.

**Jussiaeeae.** Jussiaeeae include the single genus *Ludwigia,* with about 80 species. The species

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TABLE 1. Flavonoids in the Onagraceae.[a]

| Flavonols | | | Glycoflavones | Flavones |
|---|---|---|---|---|
| kaempferol | quercetin | myricetin | vitexin | apigenin |
| 3-O-gal | 3-O-gal | 3-O-gal | isovitexin | 7-O-glu |
| 3-O-glu | 3-O-glu | 3-O-glu | orientin | 7-O-glucuronide |
| 3-O-rha | 3-O-rha | 3-O-rha | isoorientin | 7-O-glucoronide- |
| 3-O-ara | 3-O-ara | 3-O-ara | orientin-o- | sulphate |
| 3-O-rha-glu | 3-O-rha-glu | 3-O-rha-glu | acylate | luteolin |
| | 3-O-rha-gal | 3-O-diglu | vitexin-o- | 7-O-glu |
| | 3-O-diglu | 3-O-me-7-O-glu | acylate | 7-O-glucuronide |
| | 3-7-O-diglu | | lucinin | tricinin |
| | 7-O-rha | | | 7-O-glucuronide- |
| | 3-O-me, 7-O-glu | | | sulphate |
| | 3-O-xyl-gal | | | |
| | 3-O-sulphates | | | |
| | 3'-O-me | | | |
| | 3-O-glucuronide | | | |

[a] Gal = galactose, glu = glucose, rha = rhamnose, ara = arabinose, xyl = xylose.

are unevenly divided among 17 sections. As demonstrated by Eyde (1978, 1979), *Ludwigia* represents a phylogenetic line distinct from all other genera, which are, therefore, more closely related to one another than any one is to *Ludwigia.* All but a few of the species of this genus have been examined for their foliar flavonoids (Averett et al., unpubl. data).

Eight flavonoids, including three glycoflavones based on orientin and isoorientin and five flavonol 3-O-glycosides, all based on quercetin, have been identified in *Ludwigia.* Both classes of flavonoids are found among the more primitive sections *Myrtocarpus, Michelia,* and *Pterocaulon.* Only flavonols are found in sections *Tectiflora, Humboldtia, Oligospermum,* and *Caryophylloidea*; only glycoflavones are found in the remaining ten sections. Even within sect. *Pterocaulon,* only two of the five species have flavonols. Thus, with increasing specialization of the taxa, there is evidently a corresponding loss of either glycoflavones or flavonols. Overall, glycoflavones predominate. Another point to be noted is that all of the flavonols are based on quercetin and that all of the substituents are relatively simple 3-O-glycosides.

**Circaeeae.** The only genus, *Circaea,* has no clear affinities to any of the other tribes in the family. It is, however, clearly a generalized early phyletic offshoot. Previous studies (Boufford et al., 1978) reported the presence of glycoflavones and the possible presence of flavones. The earlier study was considered preliminary, was concerned with hydrolysates, and included a limited number of species. Subsequent work with additional species, especially Asian ones, has confirmed the presence of some 14 compounds among the species of *Circaea.* These include eight glycoflavones, four flavones, and two flavonols.

**Fuchsieae.** *Fuchsia* is the only genus of the tribe. Eighteen flavonoid glycosides have been identified from six ornamental species of *Fuchsia* and seven of their hybrids by Williams et al. (1983). The compounds include 11 flavonol glycosides and six flavone glycosides. Three of the six flavones are sulphates. Among the approximately 100 remaining species of *Fuchsia,* we have identified 12 flavonoid glycosides, including four flavones and eight flavonols. The flavones include two sulphates. The flavonols include six 3-O-glycosides based on kaempferol, quercetin, or myricetin; and two methyl ethers. Flavonol glycosides are found in each of the species examined. Flavones are consistently present in each of the four species of sect. *Skinnera,* occasional in the single species of sect. *Kierschlegeria,* and sporadic among one or a few species of sections *Fuchsia, Ellobium,* and *Quelusia.* The flavone sulphates are restricted to sect. *Skinnera.* With this feature, coupled with the consistent presence of other flavones, sect. *Skinnera* is the most distinct of the genus with respect to its flavonoid chemistry. This New Zealand-Tahitian group of what is otherwise an American genus is equally distinctive morphologically, although clearly a member of the genus, which is quite diverse even excluding sect. *Skinnera.*

Although both flavones and flavonols are pres-

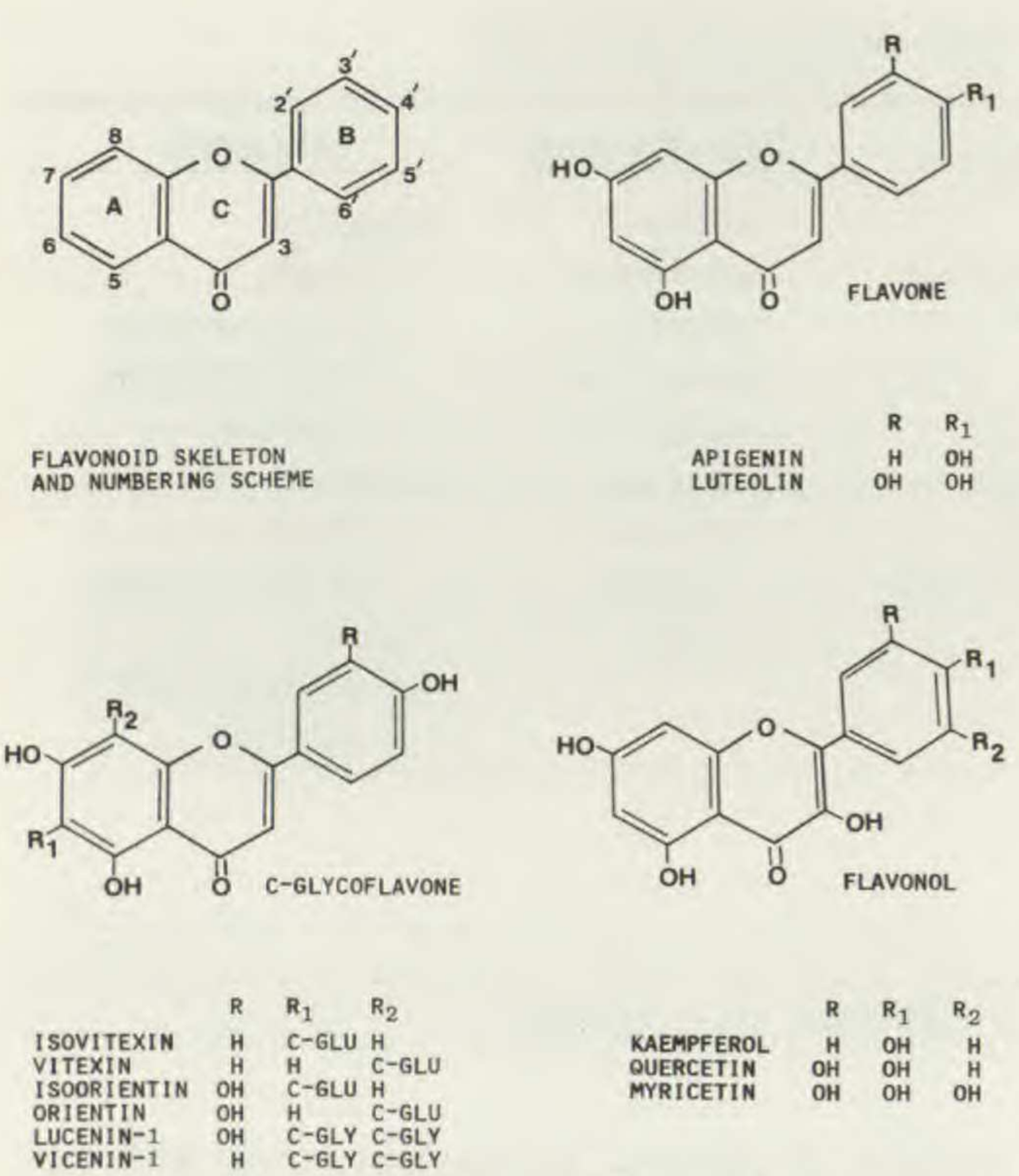


FIGURE 1. Base structures and numbering scheme for flavonoids discussed in the text.

TABLE 2. Distribution of flavonoids among the tribes of Onagraceae.

| | Flavonols | Glyco-flavones | Flavones |
|---|---|---|---|
| Tribe: | | | |
| Jussiaeeae | ● | ● | |
| Circaeeae | ● | ● | ● |
| Fuchsieae | ● | | ● |
| Lopezieae | ● | ● | |
| Hauyeae | ● | ● | |
| Onagreae | ● | ● | ● |
| Epilobieae | ● | | |

ent in *Fuchsia,* the flavones occur only in a few scattered species. In contrast to this pattern, flavonols are characteristic of the genus, occurring in every species.

**Lopezieae.** Preliminary data indicate the presence of six flavonol 3-O-glycosides and at least four glycoflavones in *Lopezia,* the only genus of the tribe. To date, glycoflavones have been detected in one of the more primitive sections of *Lopezia,* sect. *Jehlia,* and in both species of the very distinctive sect. *Riesenbachia,* which is evidently an early offshoot. Flavonols occur in both sections but are absent in one of the two species of sect. *Riesenbachia, L. riesenbachia.* Only flavonols are present in the remaining sections. Neither methylated nor sulphated flavonoids have been detected in *Lopezia.*

**Hauyeae.** *Hauya,* the single genus of the tribe, has two species. One of them, the tetraploid and hexaploid *H. elegans* DC., consists of four subspecies that range from central Mexico to Costa Rica. In all taxa of *Hauya,* four flavonol 3-O-glycosides are typically present, with an additional two occurring sporadically. In at least some populations of each of the four subspecies of *H. elegans,* however, glycoflavones are also present. The second species, the diploid *H. heydeana* Donn. Sm. & Rose, lacks glycoflavones.

**Onagreae.** Flavonoid analyses of Onagreae have been confined largely to *Oenothera,* but at least some data are available for each of the other genera (Kagan, 1967; Zinsmeister & Bartl, 1971; Zinsmeister et al., 1977; Howard et al., 1972; Averett et al., 1982; Averett & Raven, 1983a, 1983b). Only flavonols have been reported for this group but studies in progress have identified glycoflavones and flavones, as well, in a few species of *Oenothera* and *Gaura.* Some 20 compounds, including compounds with 3; 3,7; 7; and B-ring substituents, have been isolated and identified from the tribe. The substituents include monoglycosides, diglycosides, sulphates, and methyl ethers. Typically, four to six compounds are found in any one genus but these range from three in *Stenosiphon* to more than 15 in *Oenothera,* taking the genus as a whole. Additional chemosystematic analyses in the tribe are needed, especially in *Calylophus, Camissonia, Clarkia,* and *Gaura,* in view of the diversity of compounds in this tribe. Preliminary surveys both of *Gaura* and of hitherto unexamined species of *Oenothera* indicate the presence of a great diversity of systematically interesting flavonoids, both unusual flavonols and glycoflavones.

**Epilobieae.** Seven compounds, all 3-O-monoglycosides of kaempferol, quercetin, or myricetin, are found among the species of *Epilobium* and *Boisduvalia,* the two genera of this tribe. Four compounds, the 3-O-rhamnosides and 3-O-glucosides of quercetin and myricetin, are typically present in each of the sections of both genera except for the monotypic *Epilobium* sect. *Xerolobium.* In this species, myricetin 3-O-rhamnoside is absent. The additional compounds are found in *Boisduvalia* sect. *Boisduvalia* and in *Epilobium* sects. *Zauschneria* and *Chamaenerion* (Averett et al., 1978, 1979).

The lack of flavonoid diversity among so many species is striking, with only seven compounds, each a simple monoglycoside, present. Variation is largely in the presence or absence of glycosylation by an arabinosyl moiety and in the presence or absence of the single kaempferol compound. In general, the more advanced taxa have fewer compounds, representing the absence of molecules with one glycosidic substitution and/or the absence of kaempferol-based compounds. Epilobieae are highly specialized in other characteristics; the flavonoids they exhibit seem to represent an example of secondary loss of substituents; i.e., highly advanced in the terminology of Gornall and Bohm (1978). The trend is apparent both within the tribe and in comparisons between it and other tribes.

## Discussion

Harborne (1966, 1976, 1977) has considered complex glycosylation and methylation as advanced characters and the presence of glycoflavones as primitive. Gornall and Bohm (1978), although in general agreement with the primitive nature of glycoflavones, have suggested a third state, highly advanced, where substituents, once gained, are then lost. We also have suggested that a loss of substituents may be correlated with evolutionary advancement within a given taxon (Averett, 1973; Averett et al., 1978, 1979). The distribution of flavonoid types in Onagraceae (Table 2) is interesting in relation to these suggestions.

Jussiaeeae, as mentioned above, represent a distinct line within the family, a line that is characterized by the presence of glycoflavones. Flavonols are widespread in Jussiaeeae, but are relatively simple glycosides of only one base aglycone. Glycoflavones and flavonols in *Ludwigia* are found together only in the more primitive sections. The sections considered to be more specialized have lost one or the other of these two classes of flavonoids.

The rest of the family constitutes a second main line of evolution in the family. Within this line, *Circaea, Fuchsia, Hauya,* and *Lopezia*—each constituting a monogeneric tribe—seem to be relatively generalized in their characteristics, Onagreae and Epilobieae more specialized. *Lopezia* and *Hauya* have both glycoflavones and flavonols. In *Lopezia,* however, glycoflavones are restricted to a few of the most generalized taxa, apparently having been lost during the course of evolution of the genus. Flavonols, on the other hand, occur in every species examined of both *Lopezia* and *Hauya* except for one species of *Lopezia, L. riesenbachia,* which is obviously specialized. Thus, flavonols clearly predominate in these two genera, but the ones that are present are generally simple in structure, and glycoflavones are frequent in the generalized species.

*Fuchsia* has both flavonols and flavones but no glycoflavones, whereas *Circaea* has all three classes of compounds. In contrast, the two more specialized tribes, Onagreae and Epilobieae, differ markedly in their flavonoid constituents. Onagreae, like *Circaea,* have glycoflavones, flavones, and flavonols, whereas Epilobieae have the least diverse pattern of any tribe, having only flavonol 3-O-monoglycosides.

If the presence of glycoflavones is a primitive characteristic, as Harborne (1976) has postulated, the fact that this class of compounds occurs in all groups except *Fuchsia* and Epilobieae is notable. Epilobieae is a group that is clearly advanced in many of its characteristics, and one might assume that glycoflavones have been lost in the evolution of *Fuchsia* also.

Especially noteworthy in considerations of flavonoid evolution in Onagraceae is the presence of flavones in *Circaea, Fuchsia,* and the tribe Onagreae. Extensive surveys of flavonoids have not been completed in other families of the order Myrtales, but thus far, flavones have not been detected in any family other than Onagraceae (Gornall et al., 1979; Dahlgren & Thorne, 1984). This pattern suggests that ability to synthesize flavones may have evolved within Onagraceae, possibly in the ancestor of the "non-*Ludwigia*" branch of the family. If this hypothesis is correct, flavones were subsequently lost in most species of the family, including all of *Hauya, Lopezia,* and the tribe Epilobieae. It seems very unlikely that the biochemical pathway leading to the synthesis of flavones originated independently in the three groups in which they occur. Despite this, it also seems unlikely that these groups could share an immediately common ancestor in view of the major differences between them. Thus, if it can be interpreted properly, the occurrence of flavones in these three tribes might offer valuable clues to relationships within the family.

Overall, the pattern of distribution of glycoflavones and flavones in Onagraceae tends weakly to support the hypothesis that *Fuchsia* and *Circaea* might be early offshoots of the phylogenetic line leading to the tribe Onagreae, and that *Hauya, Lopezia,* and the tribe Epilobieae might not belong to this same line. Furthermore, there is some suggestion in its diversity of compounds that the tribe Onagreae might be heterogenous in origin, and new detailed information about the distribution of flavonoids in the species and genera that make up this large tribe might be helpful in evaluating this possibility.

Summarizing for the family as a whole (Table 2), glycoflavones characterize the more primitive elements but are occasionally present in all tribes except Fuchsieae and Epilobieae. Onagreae exhibit the greatest diversity of compounds, including flavonol 7-O-glycosides and B-ring substitutions. The Epilobieae exhibit the least diverse pattern, having only flavonol 3-O-monoglycosides. If these data represent, as indeed we believe they do, a proliferation of compounds during the course of evolution of the family with a reduction of compounds with further advancement, they are in general agreement with assessments of relative evolutionary states of these taxa derived from other lines of investigation. In context, however, the lack of glycoflavones in *Fuchsia* is unexpected in view of the ubiquitous distribution of this class of compounds among all other genera of the family with relatively primitive characteristics.

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